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Contents

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Effects of Antibacterial and Antifungal Agents in Diets for Laying and Breeding Hens

C. W. Carlson[1]

Numerous workers have reported on the relative ineffectiveness of low levels of antibiotics (up to 20 grams per ton) in improving the reproductive performance of chickens (1, 2, 7, 11, 13, 18, 19, 23). On the other hand, several reports have been made which indicate that low levels were effective under the conditions reported in improving the reproductive performance of laying hens (1, 6, 9, 12, 20). Higher levels of antibiotics (100 to 200 grams per ton) have been reported to more consistently improve egg production, being most effective where poorer production was prevalent (6, 14, 17, 22, 26). These studies are summarized in the Appendix.

The effects reported from the use of the arsonic acids (arsanilic acid and 3-nitro 4 hydroxyphenylarsonic acid) in diets for laying hens have been summarized by the author (4). In general, the improvement with their use has been observed as a slight increased rate of egg production and/or improved feed efficiency. Detrimental effects were reported in one instance with each of the arsonic acids (15) when fed in exceptionally high energy diets.

On the average, however, about a 3% improvement in egg production has been observed with the arsonic acids when used at the levels of 45 and 90 grams per ton (45 for 3-nitro, 90 for arsanilic acid). Dean and Stephenson (8) reported improved egg production with furazolidone (used at levels from 12½ to 50 grams per ton) and an even greater effect with the combination of furazolidone (at 15 grams per ton) and arsanilic acid (at 90 grams per ton). Data obtained over the past 2 years at this laboratory with the use of several antibiotics including penicillin, oleandomycin, oxytetracycline, chlortetracycline, and streptomycin plus arsanilic acid, the nitrofurans (furazolidone and nitrofurazone) and nystatin in diets for laying and breeding hens will be reported.

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Experimental

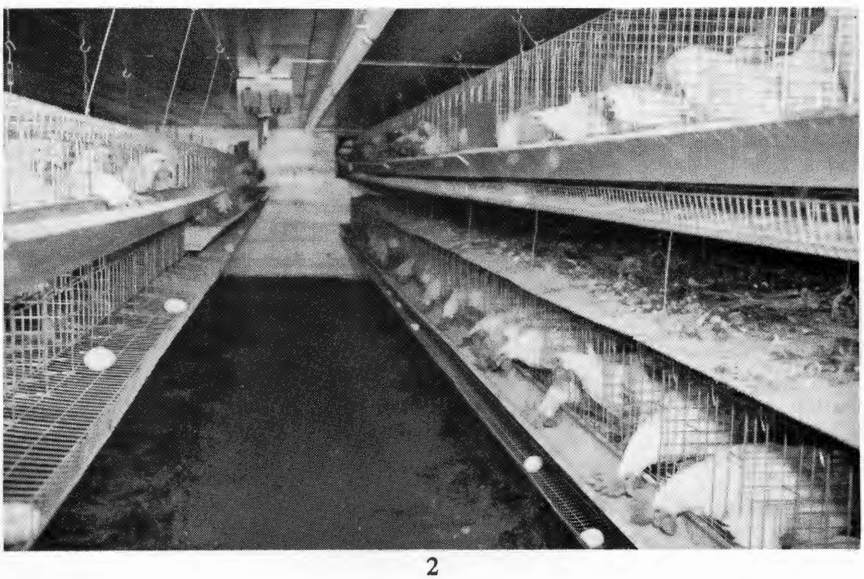
For the major part of this work and unless otherwise specified, Single Comb White Leghorn (S.C. W.L.) hens of the station flock or Leghorn-type commercial hybrids were used. Replicate groups of 40 to 60 hens were used in floor pens located for the most part under one roof—a 30- x 120-foot gable-type building of cold wall construction with provisions for twenty-four 10- x 12-foot laying pens.

In four of the five experiments, replicate groups of 15-18 hens were used concurrently with stock and experimental variables similar to the stock housed in floor pens. The replicate groups were kept in individual or group cages located in a windowless 32- x 50-foot insulated and forced ventilated building. Stocks of similar breeding, age, and rearing were used to compare the effects of environment on the responses obtained with the various experimental treatments.

The hens in floor pens were subject to greater temperature fluctuations—below freezing temperatures were common in January and February. Straw was used for litter and was replaced frequently. The hens in cages were subject to more moderate temperature fluctuations; the temperature never went below 45° F. during winter but occasionally went above 95° F. in summer. Detailed data appear in Rubida's M.S. thesis (21). The ventilation system for this house was converted from a simple 2-stage fan system to a more complex system with the use of air conditioners and a heat pump to maintain a constant year around 60-70 degree temperature for the latter part of Experiment 4.

Experiments 1, 3, and 5 were conducted for a 9-month period, start-

Individual cages used in some of the studies.



ing October 1, 1956, for Experiment 1, and October 1, 1957, for Experiments 3 and 5. The treatments began after the pullets had been in good production about 1 month, and continued through June 30 of each respective period.

The pullets for Experiments 1 and 3 had been reared in confinement to 8 weeks of age, but were grown to near sexual maturity on a green range. All pullets received the same starting and growing diets to sexual maturity. These diets contained procaine penicillin at a 4 grams per ton level. After being started on litter, the pullets for Experiment 5 were grown in group cages from 8 to 20 weeks of age. They had also received procaine penicillin at the 4 grams per ton level to 20 weeks of age.

Experiments 2 and 4 were life cycle studies in that the treatments were initiated at 1 day of age and continued through a 10-month production period, Experiment 2 in 1956-57 and Experiment 4 in 1957-58. These pullets were reared to maturity in confinement on litter, their laying cycle being about 3 months behind the hens of Experiments 1, 3, and 5.

All-mash laying diets were used, being the same for both the floor pen and cage housed hens. A 15% protein diet was used in Experiments 1, 3, 4, and 5 and a 12% protein diet was used in Experiment 2 as described in table 1. A separate study (24) conducted concurrently with Experiment 1, showed 15% protien to be adequate for maximum egg production and feed efficiency

Table 1. Formulae of Diets Used for Laying Hens

Ingredients	Diet 213, %	Diet 215, %
Gr. yellow corn	74	81
Soybean meal (44% pro.)	10	8
Meat scraps	5	
Alfalfa meal	2	2
Dried buttermilk	2	2
Steamed bonemeal	2	2
Ground limestone	3	3
Fish Meal	1	1
Salt mix*	0.5	0.5
Vitamin supplement	0.5†‡	0.5†
Calculated protein, %	15.1	12.3
Calculated productive energy, Cal./lb.	970	1003
Calculated Metabolizable energy, Cal./lb.	1358	1403

*A mixture of iodized salt containing 2½% manganese sulfate.

†To supply per pound, 1800 I. U. Vitamin A, 625 I.C.U. Vitamin D, 1 mg. riboflavin, 1 mg. pantothenic acid, 12 mg. niacin, 52 mg. choline, 4.5 mcg. cyanocobalamine, and 10 I. U. Vitamin E (used in Experiments 1, 2, and 4).

‡To supply per pound, 1800 I. U. Vitamin D, 625 I.C.U. Vitamin A, 1 mg. riboflavin, 1 mg. calcium pantothenate, 6 mg. niacin, 4 mcg. cyanocobalamine, and 5 I.U. Vitamin E (used in Experiments 3 and 5).

for caged hens under the conditions of this study. Actually, increasing the protein level to 18% through the use of a greater proportion of soybean meal and less corn resulted in poorer production and feed efficiency. With the addition of more energy to the 18% protein diet (by adding 5% stabilized yellow grease), egg production and feed efficiency were identical to that on the 15% protein diet.

Data were obtained on body weight, mortality, feed consumption, egg production, egg quality, and egg weight. For the groups of hens in floor pens, hatchability data and progeny growth data on various diets were also obtained. Pertinent data will be presented herewith. The various treatments and plans of the experiments will be given along with the results. Where data were treated statistically, an analysis of variance was conducted and calculations were made for the L.S.D.

Results and Discussion

EXPERIMENT 1

In this experiment, procaine penicillin at 4 grams per ton was included in the basal diet. Previous work (6) had indicated that penicillin elicited a positive response in improved egg production. Because of its low cost it was shown to be an economical addition to laying and breeding hen diets. There was the possibility that further additions of other antibacterial agents could be even more beneficial and therefore were used as shown in tables 2 and 4.

Group cages used in a portion of these studies, located in a windowless house.

The data on egg production show that the S.C.W.L. hens (table 2, trials 1 and 2) did not respond to any of the treatments, either the hens in floor pens or those in cages. The S.C.W.L. hens in cages (trial 2) receiving chlortetracycline produced fewer eggs than their corresponding controls. This was also true for the White Plymouth Rock (W.P.R.) hens in cages receiving oxytetracycline (table 4, trial 5), The W.P.R. hens in floor pens (trial 4) with respect to egg production did not respond significantly to oxytetracycline, although mortality was extremely low in the supplemented groups.

The commercial Hybrid A hens in cages (table 2, trial 3) seemed to respond in egg production to the nitrofurans. These differences were not significant though they approached significance at the 10% level of probability. Perhaps greater numbers would have established this observation. Mortality was also much lower in these groups; perhaps a more correct statement is that it was excessive among the Hybrid A control hens.

All of the supplemented groups of S.C.W.L. hens in floor pens produced eggs more efficiently than their corresponding controls, as was the case with the Hybrid A hens in cages. With the exception of the W.P.R. hens in cages, all of the supplements evidenced less mortality. None of the supplements appeared to have any effect upon hatchability of fertile eggs. The apparent effect of the combination of furazolidone and nitrofurazone in improving fertility is probably not real.

There were no consistent effects of these various supplements upon progeny growth (tables 3 and 5), either as given to the dams or to the progeny itself. Though furazolidone in the starter diet appeared to be detrimental to chick growth to 8 weeks of age, the pullets had largely overcome this disadvantage at 12 weeks (table 3, trial **b**). The control W.P.R. progeny of trial **d** did appear to respond to oxytetracycline, i.e. 25 grams at 4 weeks of age. Also progeny from these hens appeared to show a response to selenium, i.e., 31 grams; however, only the Vigofac response of 46 grams was statistically significant. The Barred Plymouth Rock-S.D. 21 In-

Table 2. Furazolidone, Nitrofurazone, and Chlortetracycline for a Laying Hen Diet (Experiment 1 — Trials 1, 2, and 3)

	Treatments			
Stock and Criteria	Control*	Furazolidone, 25 gm./T.	Furazolidone & Nitrofurazone, 15 gm./T.	Chlortetracycline 20 gm./T.
Trial 1				
S.C.W.L. (Floor pens — 45 hens per group, replicated)				
Egg production				
Hen Day, % (9 mo.)	56.7	57.5	57.2	56.2
Lbs. Feed/Doz. Eggs	6.6	6.1	6.3	6.1
Mortality, %	18.7	19.8	17.8	10.5
Hatchability of:†				
Fertile Eggs, %	85.7	81.4	84.4	84.0
All Eggs, %	67.3	66.5	72.8	69.0
Trial 2				
S.C.W.L. (Caged hens — 15 per group, replicated)				
Egg Production, % (9 mo.)	59.3	61.0	58.8	55.8
Lbs. Feed/Doz. Eggs	5.9	6.0	6.0	6.1
Mortality, %	10.0	13.3	13.3	6.7
Trial 3				
Hybrid A (Caged hens — 15 per group, replicated)				
Egg Production, % (9 mo.)	60.7	63.6	63.4	60.1
Lbs. Feed/Doz. Eggs	5.4	5.2	4.8	5.2
Mortality, %	33.3	6.7	6.7	10.0

*Diet 213. All diets contained 2 mg. procaine penicillin/lb.
†Approximately 700 eggs set for each treatment.

bred (B.P.R.) X S.C.W.L. (X-Bred) progeny (trial **b**) were used along with imported S.C.W.L. and W.P.R. stock in making up the various stocks for Experiment 4, to be discussed later.

An examination of the data shown in table 6 indicates that there were no consistent effects of antibacterial agents upon any of the egg quality measurements, with the possible exception of egg shell

Table 3. Effects on Progeny Growth of Furazolidone, Nitrofurazone, and Chlortetracycline in Diets of Laying Hens and Effects of Supplements Fed to Progeny (Experiment 1, Trial 1)

	Treatments			
Stock and Criteria	Control*	Furazolidone, 25 gm./T.	Furazolidone & Nitrofuranzone, 15 gm./T.	Chlortetracycline, 20 gm./T.
Trial a — females only (Feb. hatch) wt. in lbs. at 8 weeks				
Diet 109†	1.31	1.27	1.32	1.35
Trial b—females only (X-Bred, June hatch) wt. in lb. at 8 and 12 weeks‡				
Control§	1.49 (2.29)	1.46 (2.21)	1.42 (2.27)	1.45 (2.30)
Arsanilic Acid 90 gm./ton	1.43 (2.42)	1.42 (2.39)	1.35 (2.27)	1.40 (2.32)
Furazolidone, 12.5 gm./ton	1.28 (2.20)	1.32 (2.25)	1.25 (2.14)	1.32 (2.20)
Arsanilic Acid & Furazolidone	1.43 (2.27)	1.45 (2.36)	1.42 (2.21)	1.46 (2.31)

*Diet 213. All diets contained 2 mg. procaine penicillin/lb.

†Replacement stock, 50+ pullets from each floor pen fed on Diet 109 containing 62% yellow corn, 5% meat scraps, 1% fish meal, 2% alfalfa meal, 25% soybean meal, 2% steamed bonemeal, 2% dried buttermilk, ½% salt mix, and vitamins to supply, per lb.: 1800 I.U. Vit. A, 625 I.C.U. Vit. D, 2 mg. riboflavin, 2 mg. pantothenic acid, 12 mg. niacin, 52 mg. choline, 2 mcg. Vit. B_{12}, and 4 mg. penicillin.

‡12-week weight in parentheses.

§Replacement stock on life cycle study, approximately 12 pullets from each floor pen. Diet identical to above except no penicillin was added, and 10 I.U. of Vit. E per lb. was used.

Table 4. Oxytetracycline in a Diet for White Plymouth Rock Laying Hens (Experiment 1, Trials 4 and 5)

	Trial 4—Floor pens*		Trial 5—Cages†	
	Control‡	Oxytetracycline, 20 gm./T.	Control‡	Oxytetracycline, 20 gm./T.
Egg Production, Hen-Day, % (9 mo.)	48.7	50.4	53.6	47.9
Lbs. Feed/Doz. Eggs	6.7	6.6	6.6	6.6
Mortality, %	8.0	2.6	6.7	13.3
Hatchability of:				
Fertile Eggs, %	79.4	78.3	——	——
All Eggs (1000 + set), %	63.7	64.1	——	——

*60 per group, replicated.

†15 per group in individual cages, replicated.

‡Diet 213. All diets contained 2 mg. procaine penicillin/lb.

thickness. The tetracycline antibiotics did seem to cause the production of a slightly thicker egg shell in four of the five trials. This would be in line with field reports that the use of these antibiotics in feeds has effectively reduced egg breakage. A thicker shell should be a stronger egg shell, although that is not always true.

Table 5. Effect on Progeny Growth of Oxytetracycline in a Diet for White Plymouth Rock Laying Hens (Experiment 1, Trial 4)

	Floor Pens*	
	Control†	Oxytetracycline, 20 gm./T.
Trial c—Males, 3 groups of 5 chicks from each floor pen (Wt. in gm. at 4 wks.)		
Control‡	366	365
1% Vigofac	354	357
Oxytetracycline, 20 gm/T.	372	357
Diet 109§	273	252
Trial c—Females, 60 + chicks from each floor pen (Wt. in lbs. at 8 wks.)		
Diet 109§	1.43	1.38
Trial d—Replicate groups of 10 or more chicks from each floor pen (Wt. in gm. of males + females ÷ 2 at 4 wks.)		
Control‡	335	352
Reserpine, 2mg./lb.	358	351
½% Vigofac	381	360
Selenium, 2 ppm	366	357
Oxytetracycline, 20 gm./T.	360	357

*60 per group, replicated.
†Diet 215. All diets contained 2 mg. procaine penicillin/lb.
‡Containing 53% gr. yellow corn, 36% soybean meal (50% protein), 5% yellow grease, 3% steamed bonemeal, ½% limestone, ½% salt mix, 2% alfalfa meal, and to supply per lb.: 1800 I.U. Vit. A, 625 I.C.U. Vit. D, 2 mg. riboflavin, 2 mg. pantothenic acid, 12 mg. niacin, 52 mg. choline, 2 mcg. Vit. B_{12}, and 4 mg. penicillin.
§See Table 3 for composition of Diet 109.

In general, one would have to conclude that although there were no real responses elicited by the various maternal dietary supplements in this experiment, there were some indications of possible responses and further work would be necessary to confirm these observations.

EXPERIMENT 2

This was a life cycle experiment involving the use of penicillin or arsanilic acid alone and in combination, throughout the life of the X-Bred and commercial Hybrid A pullets used. The stock was reared in confinement and was approximately 3 months younger than the stock used in Experiment 1. Although the hybrid was of the same breeding, the performances of the two groups need not necessarily be comparable due to seasonal environmental differences.

The starter diet used to 8 weeks of age was identical to the starter Diet 109 described in table 2, except that no antibiotic was used in the basal, and the vitamin E supplement was not used. At 8 weeks of age the protein content of the diet was reduced to 16% by replacing soybean meal with corn, and at 16 weeks of age, Diet 215 was used—a 12% all-mash diet.

At no time during the course of growth of these pullets was there any evidence of a growth response to either arsanilic acid or penicillin.

Where penicillin had elicited growth responses as great as 49% in batteries at this laboratory in the winter of 1951 (3), in this later study there was no response—typical of what many workers have reported concerning the loss of apparent effectiveness of the antibiotics.

The results obtained from this experiment during the 10 months of production are given in table 7.

Table 6. Egg Quality Summary* (Experiment 1)

	Treatment				
Stock & Criteria	Control (Diet 213)	Furazolidone, 25 gm./T.	Nitrofurazone & Furazolidone, 15 gm./T.	Chlortetracycline 20 gm./T.	Oxytetracycline, 20 gm./T.
S.C.W.L. - Floor pens - (Trial 1)					
Haugh Unit†	79	78	78	78	——
% Blood spots ‡	12	14	16	18	——
Yolk color§	16.0	16.0	16.0	16.3	——
Egg Wt., oz./doz.	25.6	25.1	25.1	26.1	——
Shell thickness, in.	.021	.021	.021	.022	——
S.C.W.L. - Cages (Trial 2)					
Haugh Unit	78	78	78	76	——
% Blood spots	22	13	22	11	——
Yolk color	15.5	16.0	15.8	16.1	——
Egg Wt. oz./doz.	26.8	25.8	26.2	25.9	——
Shell thickness, in.	.021	.021	.021	.022	——
Hybrid A - Cages (Trial 3)					
Haugh Unit	81	80	81	81	——
% Blood spots	10	18	13	13	——
Yolk color	16.5	16.3	16.4	16.4	——
Egg Wt., oz./doz.	28.4	27.0	27.4	27.0	——
Shell thickness, in.	.020	.021	.020	.021	——
W.P.R. - Floor (Trial 4)					
Haugh Unit	79	——	——	——	76
% Blood spots	8	——	——	——	5
Yolk color	15.2	——	——	——	15.1
Egg Wt., oz./doz.	26.9	——	——	——	27.6
Shell thickness, in.	.020	——	——	——	.020
W.P.R. - Cages (Trial 5)					
Haugh Unit	75	——	——	——	81
% Blood spots	10	——	——	——	17
Yolk color	15.9	——	——	——	15.5
Egg Wt., oz./doz.	26.3	——	——	——	26.3
Shell thickness, in.	.0190	——	——	——	.0193

*Average of 4 breakout periods, 1 egg/hen, Jan., March, April, and June, yolk colors for April and June only.

†According to R. R. Haugh (10) - a figure calculated on the basis of the relationship of albumen height to egg weight.

‡Every visible blood spot was recorded, see picture for mirror viewers.

§As determined by direct comparison of colors on the Heiman-Carver color rotor.

These pullets were extremely slow in coming into production, requiring from 30-50 days longer in reaching 50% production than the same types would normally require. It seems quite evident that the 12% protein diet used was not adequate for maximum egg production with this stock and under these conditions. That may be one explanation for the variations in rates of egg production encountered in this experiment with the X-Bred hens. It is apparent that the X-Bred stock did not respond to the various treatments; actually there may have been some detrimental effects of either supplement alone (trial 1). The combination appeared to bring the performance up to that of the control groups. None of these differences were significant, however.

The Hybrid A hens did appear to respond to penicillin (trial 2), either with respect to egg production or the resulting feed efficiency. The data for trapnested egg pro-

Table 7. Arsanilic Acid and Penicillin Additions to a 12% Protein Diet (Experiment 2)

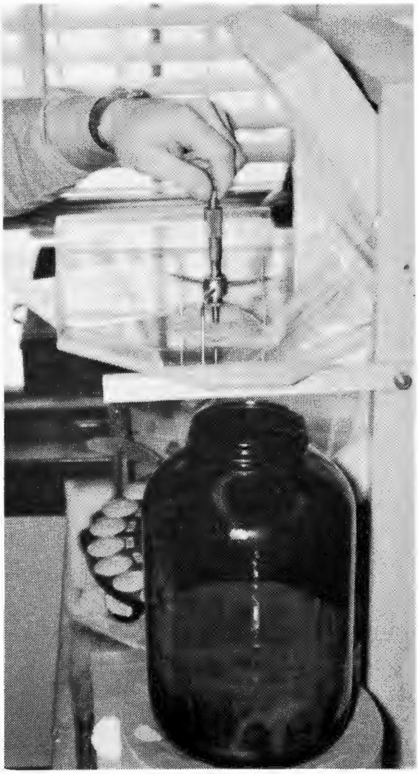
	Treatments			
Stock-Criteria	Control (Diet 215)	Arsanilic Acid 90 gm./T.	Penicillin, 4 gm./T.	Arsanilic Acid and Penicillin
X-Breds - (Trial 1)				
Days to 50% Prod.	197	204	220	200
Egg Prod. (Hen-Day), %	57.4	54.5	52.4	56.8
No. Trapnested eggs*	63.6	60.3	64.3	59.9
Lbs. Feed/Doz. Eggs	4.7	5.4	5.4	5.3
Mortality, %	9.1	9.1	9.1	5.5
Haugh Unit†	86	85	85	85
Blood Spots, %	5	3	3	6
Yolk Color	15.8	15.8	15.9	15.8
Egg Weight, oz./doz.	24.0	23.2	23.8	24.0
Shell Thickness, in.	.020	.020	.021	.020
Hybrid A - (Trial 2)				
Days to 50% Prod.	206	214	219	198
Egg Prod. (Hen-Day), %	51.8	50.7	56.5	56.1
No. Trapnested Eggs*	59.5	58.8	62.2	67.6‡
Lbs. Feed/Doz. Eggs	5.4	5.5	5.0	4.9
Mortality, %	10.9	10.9	7.3	14.6
Haugh Unit†	91	92	91	89
Blood Spots, %	10	6	7	6
Yolk Color	14.6	15.4	14.7	15.2
Egg Weight, oz./doz.	24.5	25.5	24.6	24.8
Shell Thickness, in.	.019	.019	.019	.019

*Average of all hens surviving the 10 month experiment—trapnested 3 days per week.
†All egg quality measurements are the average of breakouts in February, April, and June, one egg from each hen in production, yolk colors in April and June only.
‡Significantly different than the control pen at the 5% level of probability.

duction indicate that the combination of both supplements was necessary to elicit a significant response.

The supplements did not show any consistent effects upon mortality, Haugh Unit (10) values, incidence of blood spots, egg weight, or shell thickness. Arsanilic acid seemed to cause the Hybrid A hens to produce eggs with a slightly darker yolk color. One point on the Heiman-Carver yolk color scale normally would not be detectable by candling the unbroken egg, however there have been field reports indicating that arsanilic acid will cause the production of darker yolks. Arsanilic acid did not influence incidence of blood spots.

Mirror viewers and Heiman-Carver Color Rotor



Measuring the albumin height for the Haugh Unit determinations.

EXPERIMENT 3

The plan and summary of results obtained in this experiment are given in tables 8 (trials 1 and 2) and 10 (trials 3 and 4). The trials were run concurrently and the penicillin treatment in trials 1 and 2 was identical to the control treatment of trials 3 and 4.

All of the treatments in the group cages reduced egg production about 5% and feed efficiency about 0.4 pound. The penicillin and the higher level of oleandomycin also seemed to accentuate the already high mortality. A high percentage of this mortality was due to cannibalism, mostly "blowouts," so called, with that condition resulting in "pickouts." These hens had been severely debeaked in an attempt to correct this condition, but to little avail. Group cages would not seem to be adaptable to this strain of S.C.W.L. stock, whereas another strain of S.C.W.L. hens was subsequently used in the same facilities with very little mortality.

Oleandomycin apparently was also detrimental to the floor pen

Table 8. Oleandomycin and Penicillin Additions to a Diet for Laying Hens (Experiment 3, Trials 1 and 2)

	Treatments			
	Control	Oleandomycin		Penicillin
Criteria	(Diet 213)	2 gm./T.	10 gm./T.	4&25 gm./T.*
Trial 1				
S.C.W.L.—18 per group, in group cages, replicated				
Egg Prod. (Hen-Day), %				
Oct. 1 to Jan. 1	54.1	54.2	52.9	50.8
Oct. 1 to May 15	53.0	47.7	48.9	48.4
Lbs. Feed/Doz. Eggs	6.2	6.6	7.0	6.6
Mortality, %	35.0	30.6	42.8	66.4
Trial 2				
S.C.W.L.—40 per group, in floor pens, replicated				
Egg Production:				
% Oct. 1 to Jan. 1	68.8	70.9	71.4	65.8
No. eggs to Jan. 1†	26.5	27.7	26.5	26.9
% to July 1	59.2	60.4	60.0	53.9
No. eggs to July 1†	65.1	72.9‡	68.3	65.8
Feed Efficiency:				
Lbs. Feed/Doz. Eggs				
Oct. to Jan. 1	4.5	4.3	4.1	4.6
Oct. to July 1	5.1	5.0	4.8	5.4
Mortality, %	18.7	16.3	26.3	27.5
Hatchability of Fertile Eggs, %	73.1	69.7	66.1	71.8
Fertility, % (1000+eggs)	91 (90-93)§	83 (76-90)	83 (79-87)	83 (74-92)
Hatchability of all eggs, %	66.2	57.4	54.8	59.3
Egg Quality‖				
Haugh Units	73	74	72	74
Wt., oz./doz	24.4	24.0	24.2	24.2
Shell Thickness—in.				
Feb.	.018	.019	.019	.020
May	.015	.016	.015	.016
June	.016	.016	.016	.016

*4 gm./ton to January 1, 25 gm./ton from January 1.
†Average of individual trapnest records, 3 days per week, of hens surviving to July 1.
‡Significant at the 5% level of probability.
§Variation between replicate groups.
‖Average of three breakouts, one egg from each hen, Feb., May, and June.

hens which died, in that the numbers of eggs laid by the survivors which received oleandomycin were greater than those laid by the similar controls, whereas the percentage production data are nearly identical. The hens that died would have had to have laid at a poorer rate than the controls to balance off the superior performance of the survivors. This same effect seemed apparent for those hens in floor pens that received penicillin.

That the 2 gram level of oleandomycin did improve the performance of the survivors in floor pens is evidenced by the significant average of 7.8 more eggs laid per bird. This level of oleandomycin was apparently more effective than the higher level, and is presently being used in another trial. The hens receiving the higher level of oleandomycin and the penicillin treatments again showed the higher mortality rates. What this mortality was due to was not ascertained. No particular disease outbreak was evident nor diagnosed, although there were only a few sporadic post mortems conducted.

This was the first time at this station, where large numbers of eggs were set, that antibiotic treatments apparently showed a consistent detrimental effect upon fertility. It is not likely that this difference was due to diets but was probably due to inherent male or pen differences. The variation between replicate groups indicates that this was true, i.e. one of the supplemented replicate groups was as good as the control groups.

There were no effects upon egg quality noted in this trial as indicated in table 8 for Haugh Unit value, egg weight, or shell thickness. Although the data are not presented, there were no differences in blood spot incidence or yolk color nor in maintenance of body weight.

Growth of progeny was not greatly influenced by maternal diet. Though the control progeny showed a slightly slower growth rate in trial **a**, this was not evident in the supplemented groups of trial **a** nor in the control groups of trial **c** (table 9).

None of the antibiotic supplements fed to the progeny in these trials improved growth in all instances. Penicillin definitely showed no growth response, whereas Pro-Strep[2] evidenced some response in six out of eight instances, and oleandomycin evidenced some response in seven out of eight instances. Erythromycin (trial **c**) was not as effective as reported by McGinnis and others (16). Neovite, Vigofac, and Omafac, fermentation products of commercial source reputed to contain unidentified factors, all showed growth responses, whereas Dynasol 80-S, a commercially prepared fish solubles concentrate on soybean meal, was only partially effective in improving growth. At the level used, the tranquilizer reserpine retarded growth in this instance.

Streptomycin added to the penicillin already in the diet evidenced some improvement in egg produc-

[2]An antibiotic supplement containing 5 grams of penicillin and 15 grams of streptomycin per pound.

tion to January 1 with both the W.P.R. and New Hampshire (N.H.) hens — trials 3 and 4 (table 10). After that time, the performance of the W.P.R. hens dropped off but was not affected by treatment. Similarly, but to a lesser degree, the control group of N.H. hens also dropped off in performance. However in this case, the hens receiving streptomycin more nearly maintained their previous rate of production.

The resulting difference between groups was significant. Feed efficiency was also improved in this part of the trial. Those hens receiving streptomycin showed less mortality in both trials. In the latter phase of the trials, the hens receiving streptomycin along with penicillin were essentially receiving

Tabe 9. Effects on Progeny Growth of Oleandomycin and Penicillin Additions to a Diet for Laying Hens and Effects of Other Additions to Progeny Diet (Experiment 3, Trial 2)

	Treatments			
Criteria	Control (Diet 213)	Oleandomycin 2 gm./T.	Oleandomycin 10 gm./T.	Penicillin, 4 & 25 gm./T.*
Trial a—January Hatch (Wt. in gm. at 4 wks: males + females ÷ 2; 4 groups of 4 chicks ea.)				
1. Control Diet 102†	245	265	241	269
2. 2 gm. Penicillin/T.	269	254	265	263
3. 5 lbs. Pro-Strep/T.	286	287	278	263
4. 2 gm. Oleandomycin/T.	271	286	274	282
5. As No. 2 + 1% Neovite	281	293	289	277
6. As No. 2 + ¼% Vigofac	287	284	297	272
7. As No. 2 + ½% Omafac	277	282	280	284
8. As No. 2 + 3% Dynasol 80-S	284	263	276	266
9. As No. 8 + Oleandomycin 2 gm.	297	267	278	279
10. As No. 8 + Reserpine, 2 mg./lb.	271	262	267	253
Trial b—February Hatch only (Lbs. at 8 weeks; 50 pullets/hen pen)				
Diet 109	1.28	1.35	1.37	1.32
Trial c—June Hatch (Wt. in gm. at 4 wks: males + females ÷ 2; 4 groups of 4 chicks ea.)				
Control Diet 110‡	297	302	300	305
4 gm. Penicillin/T.	280	302	307	295
5 lbs. Pro-Strep/T.	316	307	334	300
2 gm. Oleandomycin/T.	295	320	315	312
2 gm. Erythromycin/T.	308	292	304	282

*4 gm./ton to January 1, 25 gm./ton from January 1.

†Containing, in percent, gr. yellow corn 60, soybean meal (50% protein) 32, alfalfa meal 2, steamed bonemeal 3, yellow grease 1.5, ground limestone 0.5, salt mix (iodized salt in 2½% Mn SO_4) 0.5, d-1 menthionine 0.05, and per lb., 1800 I.U. Vit. A., 625 I.C.U. Vit. D, 2 mg. riboflavin, 2 mg. panththenic acid, 12 mg. niacin, 52 mg. choline, 4.5 mcg. cyanocobalamine, and 10 I.U. Vit. E.

‡Containing, in percent, gr. yellow corn 44, soybean meal (44% protein) 45, steamed bonemeal 3, yellow grease 5, limestone 0.5, salt mix 0.5, d-1 methionine 0.05, and vitamins as above.

Pro-Strep at the level of 5 pounds per ton.

There were again no differences in hatchability of fertile eggs or in the various egg quality measurements. Also as to progeny growth, there were no differences in trials **aa** or **bb** (table 11). In trial **cc**, the W.P.R. progeny from the streptomycin fed hens grew at the most rapid rate within each treatment. This does indicate the possibility of some carry-over effect due to streptomycin in the maternal diet. This was not evident earlier in this experiment, nor in previous work at this station (9).

In general, the previous observations in trials **a** and **c** were confirmed, i.e. growth responses were obtained from oleandomycin and Pro-Strep and from the fermentation products. On the other hand, Dynasol 80-S appeared more effective in this latter instance, indicating a breed difference in the response to unidentified factor sources.

EXPERIMENT 4

This was the other life cycle experiment, involving the X-Bred progeny from Experiment 1 and other imported stock. All stock was reared to 20 weeks of age on litter and in confinement and, with respect to each of the four treatments,

Table 10. Streptomycin Additions to Penicillin-Containing Diets for Laying Hens (Experiment 3, Trials 3 and 4)

Criteria	Trial 3 - W.P.R.* Control‡	Trial 3 - W.P.R.* Streptomycin§	Trial 4 - N.H.† Control‡	Trial 4 - N.H.† Streptomycin§
Egg Production:				
%, Oct. 1 to Jan. 1	62.4	65.5	53.5	55.9
No. Trapnested**	24.8	25.5	20.5	21.7
%, Jan. 1 to July 1	49.0	48.0	48.3	52.4
No. Trapnested**	37.4	38.0	36.8	41.6‖
Lbs. Feed/Doz. Eggs:				
Oct. to Jan.	5.4	5.1	5.5	5.6
Jan. to July	6.7	6.8	5.9	5.3
Mortality, %	13.6	9.6	8.5	5.0
Hatch of Fertile Eggs, %	75.6	73.4	90.5	86.1
Egg Quality,				
Haugh Units	84	83	73	74
Weight, oz./doz.	27.2	27.0	25.1	24.7
Shell Thickness, in.				
Feb.	.020	.020	.019	.021
Apr.	.015	.015	.015	.015
June	.016	.016	.016	.017

*Replicate groups of 60 hens per group in floor pens.
†60 hens per group in floor pens.
‡Diet 213 with 2 mg. procaine penicillin to Jan. 1. From Jan. 1 to July 1 the penicillin was increased to 12.5 mg./lb.
§25 mg./lb. to Jan. 1, from Jan. 1 to July 1 the level was increased to 37.5 mg./lb.
‖Significant at the 5% level of probability.
**Three days per week, survivors only.

in the same pens. As can be seen by the data for days to 50% production, each of the treatments slightly lengthened the time required to reach this stage of production. This slight disadvantage was quickly overcome, however, and at the end of 3 months, the S.C.W.L. hens from each of the treated lots had laid at a superior rate. None of the other groups showed this trend, however, except for the X-Bred caged hens receiving both supplements together with Penicillin V. This trend of the latter group persisted through the entire 10 months of production; all of the other groups showed no responses to any treatment after 7 months of production.

Penicillin V evidenced some detrimental effects upon the unsupplemented group and each group receiving the single supplement. This is suggestive that the form of penicillin is not the reason for the detrimental effects of penicillin reported in Experiment 3. Arsanilic acid and Penicillin V were not antagonistic in this latter work. No explana-

Table 11. Effects on Progeny Growth of Streptomycin Additions to Penicillin-Containing Diets of Laying Hens and Effects of Other Additions to Progeny Diet (Experiment 3, Trials 3 and 4)

	Trial 3—W.P.R.*		Trial 4—N.H.†	
Criteria	Control†	Streptomycin§	Control‡	Streptomycin§
Trial aa—January Hatch (Wt. in gm. at 4 wks: males + females ÷ 2; 4 groups of at least 4 chicks each for W.P.R., 2 groups of 6 for N.H.)				
1. Control Diet 102	279	267	272	252
2. 2 gm. Penicillin/T.	281	299	269	277
3. 5 lbs. Pro-Strep/T.	306	307	304	297
4. 2 gm. Oleandomycin/T.	297	318	307	277
5. As No. 2 + 1% Neovite	287	306	309	279
6. As No. 2 + ¼% Vigofac	311	293	338	291
7. As No. 2 + ½% Omafac	308	314	287	254
8. As No. 2 + 3% Dynasol 80-S	290	307	305	297
9. As No. 8 + Oleandomycin	328	299	295	289
10. As No. 8 + Reserpine, 2 mg./lb.	287	293	269	276
Trial bb—March Hatch (females only—wt. in lb. at 8 wks.; 40 chicks from each pen				
Diet 109	1.34	1.34	——	——
Trial cc—June Hatch (wt. in gm. at 4 wks.; males + females ÷ 2)				
Control Diet 110	329	350	295	301
4 gm. Penicillin/T.	310	321	293	290
5 lbs. Pro-Strep/T.	349	378	354	309
2 gm. Oleandomycin/T.	346	352	309	309
2 gm. Erythromycin/T.	338	350	311	308

*Replicate groups of 60 hens per group in floor pens.
†60 hens per group in floor pens.
‡Diet 213 with 2 mg. procaine penicillin to Jan. 1. From Jan. 1 to July 1 the penicillin was increased to 12.5 mg./lb.
§25 mg./lb. to Jan. 1, from Jan. 1 to July 1 the level was increased to 37.5 mg./lb.

tion can be given for the earlier reported antagonistic effect of procaine penicillin and arsanilic acid (5).

The data in table 12 show no consistent effects of the treatments upon feed efficiency or mortality. On the over-all basis, the combined treatments evidenced a slight advantage in feed efficiency and mortality, but certainly not a real difference. There were no consistent effects of the various treatments upon the hatchability of fertile eggs. The poorer results obtained from the X-Bred and W.P.R. hens given the various treatments may be due to the fewer numbers of eggs set, since the performance of the S.C. W.L. hens was remarkably uniform.

As to the various egg quality measurements, the Haugh Unit value, egg yolk color, shell thickness, or egg weight were not influenced by the various treatments. Neither was there any consistent effect of the supplements upon incidence of blood spots.

With respect to progeny growth (table 13), there did appear to be a carry-over effect to their chicks from the hens receiving supplements. This was evident in the first growth study, trial **a**, with the chicks on litter. A 10-gram difference with 150 chicks would be a highly significant difference. However this was not evident with battery brooded chicks later in the experimental period (trials **b** and **c**). There was not an opportunity to repeat the conditions of trial **a**, so it cannot be ascertained whether this effect was due to conditions brought about by growing the chicks in brooder houses on straw litter or to some other factor.

EXPERIMENT 5

This experiment was conducted concurrently with Experiment 3, but was conducted exclusively with Hybrid B hens in individual cages. An antifungal agent, nystatin, was used alone and in combination with oxytetracycline. As can be seen from the data in table 14, neither supplement alone had any marked effect upon egg production, but the combination of the two appeared to be effective in improving production, at least for the first 4 months of the experimental period. The small difference persisting to the end of the experiment was not a significant difference. However, the fact that these hens were in cages, together with the probable earlier difference, would indicate that further work with an antifungal agent might well be considered with hens in cages and in floor pens with litter.

Winans (25) made a study of the *Candida albican* population of feces from hens in this study, along with another group of hens receiving penicillin at 4 grams per ton. He observed that hens receiving penicillin had much greater numbers of the organism in their feces than control hens, whereas hens receiving either oxytetracycline or nystatin showed fewer numbers than the controls. Whether or not the detrimental effects of penicillin observed in Experiments 3 and 4 were due to this increased incidence of *Candida albicans* remains to be demonstrated by further work.

Table 12. Furazolidone and Arsanilic Acid Supplements for a Laying Hen Diet (Experiment 4)

Criteria—Stock	Control Diet 213	Furazolidone, 15 gm./T.	Arsanilic Acid, 90 gm./T.	Furazolidone & Arsanilic Acid
	Treatments			
Days to 50% Production				
S.C.W.L.—Floor*	171	174	174	175
X-Bred, W.P.R.—Floor†	169	175	176	171
X-Bred, Cages‡	175	178	176	179
X-Bred, Cages§	171	179	175	178
% Egg Production—at 3 mo. (Hen-Day basis—Cumulative)				
S.C.W.L.—Floor	65.3	72.4	69.0	70.1
X-Bred, W.P.R.—Floor	66.9	65.2	65.8	66.3
X-Bred, Cages	73.4	73.2	73.8	72.6
X-Bred, Cages§	70.2	70.6	71.0	74.0
% Egg Production at 7 mo. (Cumulative)				
S.C.W.L.—Floor	70.0	68.6	70.4	71.3
X-Bred, W.P.R.—Floor	64.5	62.5	63.2	63.2
X-Bred, Cages	69.6	69.1	69.4	68.9
X-Bred, Cages§	65.1	63.9	64.4	67.7
% Egg Production at 10 mo. (Cumulative)				
S.C.W.L.—Floor	68.3	67.6	66.8	69.0
X-Bred, W.P.R.—Floor	58.4	57.2	58.3	58.2
X-Bred, Cages	65.7	64.4	65.6	65.4
X-Bred, Cages§	61.7	59.6	60.6	63.4
Lbs. Feed/Doz. Eggs				
S.C.W.L—Floor	4.1	4.3	4.2	4.0
X-Bred, W.P.R.—Floor	4.9	5.0	5.0	4.9
X-Bred, Cages	4.6	4.8	4.6	4.2
X-Bred, Cages§	4.4	4.7	5.1	4.6
Mortality, %				
S.C.W.L.—Floor	18.3	20.0	30.0	23.3
X-Bred+WPR ÷ 2 -Floor	21.1	23.1	23.1	28.8
X-Bred, Cages	26.7	20.0	26.7	20.0
X-Bred, Cages§	30.0	36.7	16.7	16.7
Body Wt., % in Sept. as compared to Jan.				
S.C.W.L.—Floor	108	102	111	104
X-Bred+WPR ÷ 2 -Floor	110	104	110	105
X-Bred, Cages	107	111	103	107
X-Bred, Cages§	111	111	107	109
Hatchability, % of Fertile Eggs (approximately 600 and 300 fertile eggs, respectively)				
S.C.W.L—Floor	80.1	82.2	79.3	79.4
X-Bred+WPR ÷ 2 -Floor	86.5	80.8	74.1	82.6
Egg Quality—Av. of All Groups, (Av. of breakouts in Feb., Apr., June, & Sept.)				
Haugh Units	75	75	76	74
Yolk Color	15.8	16.0	15.9	16.0
Wt., oz./doz.	23.7	23.7	23.8	23.7
Shell Thickness, in.	.018	.018	.018	.018
Blood Spots, %	6.4	4.1	8.4	4.2

*60 pullets, imported as eggs—reared in confinement with all groups below.

†52 pullets, 32 X-Bred (B.P.R. X S.C.W.L.) & 20 W.P.R. pullets, the latter imported as eggs, the X-Breds as progeny from Experiment 1. Egg production data were the pen averages, all other data were the averages of the two breeds.

‡2 groups of 15 X-Bred hens in individual cages.

§2 groups of 15 hens in individual cages, 4 gm. of Penicillin V added to diets of all groups in series.

Table 13. Effects on Progeny Growth of Furazolidone and Arsanilic Acid Supplements in Laying Hen Diets and Effects of Other Additions to Progeny Diet (Experiment 4)

Criteria	Treatment: Control Diet 213	Furazolidone, 15 gm./T.	Arsanilic Acid, 15 gm./T.	Furazolidone & Arsanilic Acid
	(Weight in grams at 4 weeks)			
Trial a—in brooder houses in Jan. on straw litter (approximately 50 pullet chicks from each pen in each of 3 brooder houses)				
S.C.W.L.—Diet 109	174	188	198	184
Trial b—in battery brooders in March, pullets only, 22/group				
S.C.W.L.—Diet 109	190	190	195	196
Trial c—in battery brooders in Sept., 8-10 chicks each (males+females÷2)				
S.C.W.L.—Floor				
Control Diet*	221	248	254	258
8 lbs. Pro-Strep/T.	261	250	231	247
X-Bred—Floor				
Control Diet*	232	225	264	243
8 lbs. Pro-Strep/T.	228	257	244	233

*Similar to Diet 110, except 3% corn replaced 2% of the yellow grease, see table 9.

Table 14. Nystatin and Oxytetracycline Additions to a Laying Hen Diet (Experiment 5)

	Supplement: Control* Diet 213	Nystatin, 50 gm./T.	Oxytetracycline, 50 gm./T.	Nystatin & Oxytetracycline
% Egg Production (Hen Day—Cumulative)				
Oct.—Dec.	75.8	74.8	76.5	79.5
Oct.—Jan.	75.1	74.7	76.3	78.0
Oct.—Feb.	74.3	73.6	74.8	76.5
Oct.—Mar.	73.8	73.2	73.5	75.2
Oct.—Apr.	72.5	72.5	72.3	74.2
Oct.—May	71.5	72.2	70.4	73.3
Oct.—June	70.4	71.2	69.5	72.2
Lbs. Feed/Doz. Eggs				
Oct.—Dec.	4.1	4.4	4.4	4.2
Oct.—Apr.	4.4	4.6	4.7	4.5
Oct.—June	4.4	4.4	4.7	4.6

*Average of 2 replicate groups of 15 Hybrid B hens in individual cages.

Summary

Chlortetracycline, oxytetracycline, penicillin (V. and Procaine), streptomycin, oleandomycin, and arsanilic acid, furazolidone, nitrofurazone, and nystatin have been used in studies to determine their effectiveness in all-mash diets for laying hens.

In one trial with a 12% protein diet, penicillin at 4 grams per ton together with arsanilic acid caused an improvement in egg production. In the later studies, penicillin has shown some detrimental effects upon egg production, whereas the other supplements have shown promise as a result of their beneficial effects under some of the conditions reported.

Arsanilic acid was used at 90 grams per ton, streptomycin at 50 and 75 grams per ton, oxytetracycline at 20 and 50 grams per ton, chlortetracycline at 20 grams per ton, furazolidone at 15 and 25 grams per ton, nitrofurazone at 15 grams per ton (with furazolidone), oleandomycin at 2 and 10 grams per ton, and nystatin at 50 grams per ton. The amount, kind, and type of supplement to recommend for use may well be dependent upon each situation.

The continuous use of some broad spectrum antibiotic at a level of about 20 grams per ton or a combination of furazolidone at 15 grams per ton with arsanilic acid at 90 grams per ton would appear to be useful under many conditions. For treatment of various specific or nonspecific disease situations, higher levels of the antibiotics including a combination of penicillin and streptomycin or the nitrofurans and arsonic acids may be warranted. Further work is necessary to answer all the questions involved. Nystatin, or some antifungal agent, may be quite useful in combination with an antibiotic for improving egg production.

Where egg production is poor, large responses might be obtained from these various drugs. However, where egg production is good, e.g. averaging about 70%, it is not likely that any of the drugs or any combination will markedly influence the rate of egg production. Because of the variability encountered in egg production experiments, small differences, however real or beneficial, may often go undetected.

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Appendix

Some Reported Effects of Antibiotics Upon the Reproductive Performance of Chickens

Authors	Breed	Antibiotic	Level	Effect
Bentley & Hershberger (1)	S.C.W.L.	Bacitracin Terramycin Aureomycin Procaine penicillin	10-20gm/T.	None on hatchability, significantly improved progeny growth by Bacitracin & Aureomycin.
Berg, et al (2)	S.C.W.L.	Terramycin Aureomycin	4-10 ppm.	No consistent effect.
Carpenter, et al (7)	Buff Rocks	Aureomycin	10 ppm.	No effect.
Johnson (11)	N. Hampshire	Procaine Penicillin	10-100 mg/Kg.	No consistent effect.
Lillie & Bird (13)	R. I. Red	Aureomycin	1% Vit. B_{12}-Aureomycin Supplement	No effect
Petersen & Lampman (18)	S.C.W.L.	Procaine Penicillin Streptomycin Terramycin	9 gm./T.	No consistent effect.
Petersen, et al (19)	S.C.W.L.	Procaine Penicillin	4, 20, 100 gm/T.	No reproductive improvement—body weight gain increased.
Sunde, et al (23)	S.C.W.L.	Aureomycin	9 gm./T.	No consistent effect—(10% greater rate of prod. in 1st Exp.)
Carlson, et al (6)	W. P. Rock B. P. Rock N. Hampshire S.C.W.L. Hybrids	Procaine Penicillin Streptomycin Chlortetracycline Oxytetracycline Tetracycline	2 to 100 gm/T.	General improvement in production up to 13%, particularly when production was substandard. Higher levels were more consistent—less mortality. No effect on progeny or egg quality.

Elam, et al (9)	N.H. X S.C.W.L.	Procaine Penicillin	33 mg/Kg.	Increased egg production 12%.
Kennard & Chamberlin (12)		(Not indicated in the report)		Increased egg production 10%—improved feed efficiency 11%.
Nabor & Winter (17)	S.C.W.L.	Aureomycin Terramycin	100 gm/T.	Increased egg production 3-4%.
Reid, et al (20)	N.H. X S.C.W.L. N. Hampshire	Aureomycin Terramycin Bacitracin Streptomycin Penicillin	2½ to 400 gm/T.	Increased egg production 3-10%.
Ryan, et al (22)	Various	Aureomycin	100 gm/T.	Increased egg production 4.7%.
White-Stevens & Delappe (26)	S.C.W.L. N. Hampshire	Aureomycin	50 to 100 gm/T.	Increased egg production 0-11%.